JOURNAL OF PURE AND APPLIED MICROBIOLOGY, June 2013. Vol. 7(2), p. 1451-1454

Antimicrobial Studies of Sulphasomidine and its Comparison with Transition Metal Complexes

S.A. Iqbal[1] and E. H. El-Mosalaamy[2]

[1]Cresent College of Technology, Nabi Bagh, Karond, Bhopal - 462 038, India.
[2]Department of chemistry, King Abdul Aziz University, Jeddah, Saudi Arabia.

(Received: 18 April 2013; accepted: 30 May 2013)

Synthesis, characterization and antimicrobial studies of sulphasomidine complexes with Hg, Zn,and Ag have been studied. The conductometric studies using monovariation and Job's method of continuous variation indicate that complexes formed are of L_2M type and non-ionic except silver which form 1:1, complex. Analysis of the complex verifies the molecular formula which is supported by analytical data. Structure of the complexes proposed on the basis of stoichiometry are supported by spectral studies and particle size analysis etc. Antimicrobial activities of the complexes were found to be more as compared to drug alone.

Keywords: Sulphasomidine, antimicrobial drug, complex, transition metals.

Metal ion are required for many critical function in humans. Scarcity of some metal ions can leads to disease[1]. Well – known example can leads to pernicious anaemia resulting from iron deficiency; growth retardation arising from insufficient dietary zinc, and heart disease in infants owing to copper deficiency. The ability to recognize, to understand at the molecular level, and to the diseases caused by inadequate metal-ion function constitutes an important aspect of medicinal bioinorganic chemistry. Understanding the biochemistry and molecular biology of natural detoxification mechanisms and designing and applying ion–specific chelating agents to treat metal over -loads are the two components of a second major aspect of the new science that is evolving at the interface of bioinorganic chemistry and medicine.

Sulphonamides were the first antimicrobial agents effective against pathogenic bacterial infection. Sulphonamides may considered to be the derivative of sulphanilamide (para amino benzene sulphonamide). Sulphonamides are primarily bacteriostatic against many *Gram positive* and *Gram negative*, looking to their chemotherapeutic action, large number of sulphonamide have been synthesized by Kaushal, Iqbal and co-workers [2-15]

The discovery of cynocobalamine, (vitamin B_{12}), with the development of coordination compounds or complexes with organic ligands is developing very fast both in industry and medicine. For this reason we have under taken a systematic study to evaluate the antibacterial activity of our synthesized Ag, Hg, Zn and Co complexes as compared to sulphasomidine, the parent drug.

EXPERIMENTAL

Ligand-Metal Ratio

a) Pure drug sulphasomidine m.p. 243°C, of 0.005 M, were diluted to 100 ml ethanol as required and titrated conductometrically against metals salt at 30±1°C. Results were plotted in the form of a graph which indicate ligand metal ratio as 2:1 (L_2M) and in case of Ag as 1:1 ratio.

* To whom all correspondence should be addressed.
E-mail: iqbalospc@yahoo.com

b) Formation of 2:1 (L_2M) and 1:1 in case of Ag was further confirmed by Job's method[7] of continuous variation as modified by Turner and Anderson[8] from these values the stability constant (log k) and free energy change (-DF), were also calculated.

Synthesis of Complexes

The chemicals used in this synthesis were all of AnalaR grade, E-Merck. A weighed quantity of sulphasomidine was dissolved separately in minimum quantity of distilled water. The metal salt solution was prepared by dissolving (1 mole) separately in the same solvent. Ligand solution was added slowly with stirring into the metallic salt solution at room temperature, On refluxing the mixture for 3 hour and on cooling the complexes separated out. Which were filtered off, washed well with distilled water and finally dried in vacuum and weighed.

The elemental analyses of the isolated complexes were carried out using Coleman Analyzer at the Departmental Microanalytical Laboratory, CDRI Lucknow, India.

RESULT AND DISCUSSION

Bacteriostatic studies of sulphasomidine and their silver, mercury, zinc and cobalt complexes

Clinical medical microbiology is a science which is concerned with the isolation and identification of diseases producing micro-organism; i.e. Bacteria, Fungi, Viruses rickeitisia, and parasites and the techniques employed in the isolation and identification of the suspect organism involves the propagation on suitable primary culture media, selective isolation on special culture media, use of suitable living host material, determination of morphological and where applicable, staining characteristic of the organism, confirmation by biochemical and immunochemicals analysis, suitable animal inoculation, where applicable may be employed to determine

Table 1. Pysico-chemical Characterstics of sulphasomidine complexes.

S. No.	Complexes	Colour	Yield (%)	m.p.°C	Ligand metal ratio
1.	$(C_{12}H_{14}N_4O_2S)_2Ag$	White	66	290	1 : 1
2.	$(C_{12}H_{14}N_4O_2S)_2Zn$	White	52	272	2 : 1
3.	$(C_{12}H_{14}N_4O_2S)_2Hg$	White	71	245	2 : 1

Table 2. Elemental analysis of sulphasomidine complexes

S. No.	Formula of complexes	Mol wt (g/mole)	C %	H %	N %	S %	Metal (%)
1.	$(C_{12}H_{14}N_4O_2S)_2Ag$	663	36.12 (37.92)	3.93 (3.71)	14.22 (14.74)	8.51 (8.44)	28.99 (28.38)
2.	$(C_{12}H_{14}N_4O_2S)_2Zn$	620	45.65 (46.35)	4.70 (4.52)	17.69 (18.01)	10.03 (10.31)	11.18 (10.51)
3.	$(C_{12}H_{14}N_4O_2S)_2Hg$	758.13	37.50 (38.07)	4.10 (3.73)	14.08 (14.79)	8.09 (8,47)	26.51 (26.48)

From the synthesis and the analytical data of the complexes the above mentioned molecular formulae are confirmed.

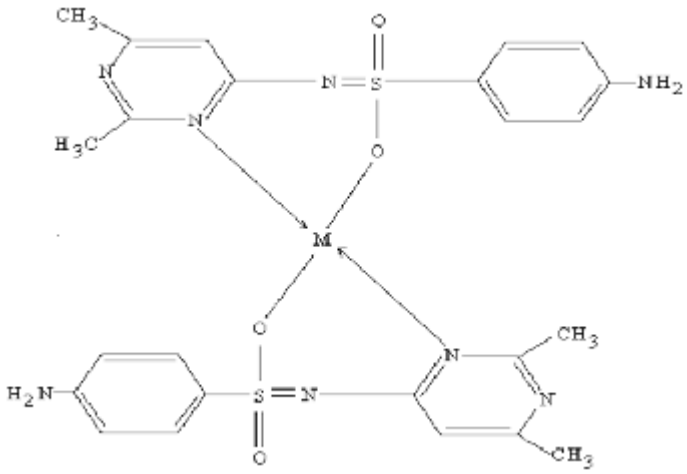
Table 3. IR studies[9-15] of sulphasomidine and their silver, mercury, zinc and cobalt complexes

Complexes	Ar-S	S=O	-C=N	SO_2NH	M-O	Heterocyclic ring
Sulphasomidine(SSD)	700s		-	3020m	-	1570m
$(C_{12}H_{14}N_4O_2S)_2Ag$	690w	1220s	1530s	3040w	670m	1580vs
$(C_{12}H_{14}N_4O_2S)_2Zn$	690s	1230m	1540s	3060w	680s	1580vs
$(C_{12}H_{14}N_4O_2S)_2Hg$	670w	1230m	1533m	3080vw	660s	1560s

Table 4. Antibacterial activity of sulphasomidine and their metal complexes

S.No.	Complexes	*Escherichia coli*	*Bacillus subtillus*
1	Sulphasomidine (SSD)	- - -	+ + +
2	SSD-Hg	+ + +	-
3	SSD-Zn	+ + +	-
4	SSD-Ag	+ + +	-

(-, +, ++, +++, ++++)
Resistant – 0-0 cm
Weakly Sensitive - 0-5 cm
Moderately Sensitive - 5-10 cm
Markedly Sensitive - 10-15 cm
Highly Sensitive - more than 15 cm



Scheme 1. Structure of the complexes of Sulphasomidine with Silver, Zinc and Mercury

pathogenicity site timing technique, instrumentation and transportation of clinical specimen are prime variables involving in the final differentiation and confirmation process.

A simple, rapid and quite useful agar streak method for measuring antibacterial activity has been described by Waksman and Reilly. The antibacterial activity of the compound is indicated by a clear or partially clear zone of inhabitation. Chick embryos have been utilized for evaluating chemotherapeutic activity; Bone marrow and tissue culture have also been used in the measurement of antibacterial activity.

The disc plate or cup plate assay methods are commonly employed for quantitative measurement of antibacterial activity. Bacteriostatic studies of sulphasomidine and their metal complexes have been carried out to see how for these complexes are active.

Preparation of solution

Stock solution of the sulphasomidine and their metal complexes were prepared by dissolving 25mg/ml of the solvent (DMF). Test solutions were prepared by appropriate dilution with hydrogen free distilled water to get desired concentrations.

Medium preparation

The glass wares were cleaned and sterilized in hot air oven at 160°C for one hour. The medium was prepared as suggested by Harper and Cawsten. Meat infusion agar was prepared by talking 500 g, of finally minced meat free from fat added in one liter of distilled water. After keeping the mixture in refrigeration for overnight, filter it through gauge piece. Subsequently, the fluid was boiled for 20 minutes and kept it overnight. Due to alteration of hemoglobin and coagulation of soluble proteins the extract becomes brown and turbid. It was, therefore, again filtered through the filter

paper, made up the volume to one liter. 10g peptone and 5g sodium chloride were added to the 1.0 liter clear solution. The mixture was heated on steam bath to get these ingredients dissolved, and P^H was a adjusted to 7.6 using normal NaOH solution. 25g agar was added and the medium was sterilized at 15lb pressure and 121°C for 25 minutes. Finally, after moderate cooling the measured volume of medium was distributed in sterile Petri dishes and left undisturbed to set at room temperature.

Preparation of antibiotic disks

For the study of antibacterial activity paper disks of appropriate diameters were used. Ten such disks were transferred to clean and dry test tube. These tubes containing the disks were dry sterilized at 140 ^{0}C. Subsequently 0.0 ml of sulphasomidine and its complexes were poured in each tube . Petri dishes containing the medium were seeded separately with fresh 18h broth culture of pathogenic organisms aseptically. After some time with the help of sterile pipette the excess of the inoculum was drained off and subsequently each plate was labeled according to the inoculating organism. With the help of a sterile fine pointed forcep, One filter paper disk soaked in the particular solution of sulphasomidine and its complexes. Finally after appropriate labeling all the inoculated organism plates with disks soaked in the solution of sulphasomidine and its complexes were incubated at 37^{0}C/ 24 h and the diameter of the zone of inhibition was measured in mm The relative degree of sensitivity was noted. During the experiment all possible precaution were taken to keep the conditions of the experiment uniform, regarding volume and thickness and the quantity of the solution used.

The diameters of zones of inhibition were measured and is recorded in Table 1.

ACKNOWLEDGMENTS

The author is thankful to the principal, Crescent College of Technology, Bhopal for providing all necessary facilities and IIT, Bombay, for providing IR spectra and x-Ray data.

REFERENCES

1. E.J ,Underwood., Trace element in human and animal nutrition 3rd ed, Academic press, New York N.Y., 1971; P-57.
2 . Farhana, Afridi, S. A. Iqbal,., and Javed, Hasan., *Orient. J. Chem*., 22(1): 195-197
3. Bal Krishanand Iqbal, S. A., *Orient. J. Chem*., 2012; **28**(4): 1883-1888.
4. P.Job, *Ann.Clin*, 1928; **113**, 10.
5. S.E.Turner and R.C. Anderson, *J Am.Chem.Soc*., 1949; **912**: 71.
6. R. Saxena., Sahdev and S. ahmed. *Orient. J. Chem*., 2010; **26**(4): 1507-1511.
7. M, Azimi, and S. Ahmed., *Orient. J. Chem*., 2011; **27**(2): 673-677.
8. C.N.R. Rao, Chemical Applications of Infra-red spectroscopy, Academic press NY., 1963
9. L.J. Bellamy. The Infra-red spectra of complex molecules.Matheun and co.Ltd. London, 1964.
10. A. Weissberger, Chem. Application of spectroscopy", Vol. XI Inter Science Publ. New York. 1956.
11. George Jacob, Syed Aftab Iqbal and E.H. El-Mossalamy, *Asian J. Chem*. 2011; 23(2): 573-576.
12. Bal krishan and Iqbal,S.A., *Orient.J.Chem*., 2012; **28**(4): 1883-1888.